LEAF CHARACTERISTICS OF SPIDER MITE RESISTANT AND SUSCEPTIBLE CULTIVARS OF PELARGONIUM X HORTORUM[1]

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Snetsinger, Balderston, and Craig (1966) reported that species and cultivars of *Pelargonium* exhibit different degrees of resistance to the twospotted spider mite, *Tetranychus urticae* Koch. MacDonald, *et al.* (1971) evaluated several methods of bioassaying host resistance and susceptibility to attack by the twospotted spider mite; they recommended a leaf disc method, because of its reliability and efficiency.

Dunnam and Clark (1938) observed that aphid populations increased indirect proportion to the number of leaf hairs on the lower leaf surface of cotton. Afzal and Ghani (1984) studied host resistance to the leafhopper, *Empoasca devastans*, as related to "toughness" and "hairiness" of two cotton varieties. In our study we have used the fecundity of adult, female, twospotted spider mites as an index of host resistance and susceptibility and attempted to relate this index to the number and kinds of leaf hairs, and thickness of the cuticular and epidermal layers.

Methods—Two pelargonium cultivars were selected—one classified as spider mite resistant and one susceptible to spider mite attack. The cultivar G54 is a seed propagated diploid developed by R. Craig and D. E. Walker, The Pennsylvania State University in 1963. Cultivar

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G10 ('King Midas') is an asexually propagated diploid developed by X-ray irradiation by E. Richards in Seattle, Washington. Our observations on live plant materials suggest the G54 is effectively resistant to spider mite attack, while G10 is relatively susceptible to such attack.

Rearing the spider mites: A stock culture of *Tetrancyhus urticae* was reared on the bean, *Phaseolus vulgaris* L. 'Bountiful'. Bean seeds were planted in vermiculite in 51 × 33 × 7 cm wooden flats in the greenhouse. The bean seeds were germinated in the greenhouse under a 24-hour photoperiod using fluorescent light to supplement natural daylight. Seedlings were watered daily and fertilized twice weekly with a 15-30-15 water soluble fertilizer at a rate of 14 g/liter of water. After two weeks, the plants were transferred, four each, to 15 × 11 × 12 cm peat pots filled with vermiculite. Young plants, at about five weeks of age, were placed among other plants infested with a culture of *T. urticae*; these were maintained in controlled environment chambers. The mite culture was an isogenic population collected from a greenhouse at University Park in 1962.

Environmental chambers (Sherer Model Cel 25-7) were used for mite cultures and experiments, and were maintained on a 12-hour photoperiod with four 25-watt incandescent bulbs and six 110-watt fluorescent lamps; day temperatures were 23±1°C and night temperatures 18±1°C; and relative humidities varied from 50 to 70% during the day and 70 to 90% at night.

Leaf disc technique: The third leaf from the apex of each experimental *Pelargonium* × *hortorum* Bailey plant was removed and taken to the laboratory. These leaves were cut into 12 mm diameter discs with a number 9 corkborer. Leaf discs were placed one each on a 33 mm filter paper contained in a 60 × 35 mm plastic, disposable microdiffusion dish.

One mature female twospotted spider mite was transferred to each leaf disc with the aid of a dissecting needle and stereoscopic microscope. Filter paper was moistened with distilled water to retain the mites on the desired leaf surface and prevent the leaf disc from desiccating. The microdiffusion dishes on a glass plate were placed in an environmental chamber, and maintained at the same photoperiods, temperatures, and relative humidities, as the stock culture.

After 72 hours, the numbers of eggs laid on each leaf disc were

counted. The number of eggs produced during this period was used as the criterion to compare resistance of the two pelargonium cultivars.

Leaf sectioning technique: A standard method of obtaining the leaf samples was used to measure cellular configuration, thickness of the cuticle and the epidermis, as well as the total thickness of leaves. The third leaf from the apex was removed and taken to the laboratory for sectioning. Five pieces (5 × 5 mm) were sliced from each leaf with a razor blade and fixed in F.A.A. (70% EtOH 90 cc, glacial acetic acid 5cc, formalin 5cc) immediately. After 24 hours, the tertiary-butyl alcohol series was used for dehydration. Both low melting point (50 to 52°C.) and high melting point (56 to 58°C) paraffins were used for infiltration, after which the leaf pieces were embedded in Paraplast. The pieces were sectioned to a thickness of ten microns, perpendicular to the vein. The sections were then affixed with Haupt's adhesive to a microscope slide. Two slides each with 60 or more sections were made from each piece of leaf. The paraplast was removed and the tissue was hydrated in a xylene-ethyl alcohol-water series and stained with Safranin O biological stain for 30 minutes. The specimens were dehydrated with a water-ethyl alcohol series and counterstained with Fast Green FCF biological stain. Cover slips were mounted on the slides . . . the leaf tissue was examined and measured under a microscope with an ocular micrometer.

The number of hairs from a 5.3 mm^2 piece of leaf was counted; two types of hairs were observed. The simple type hairs (Figure 1,

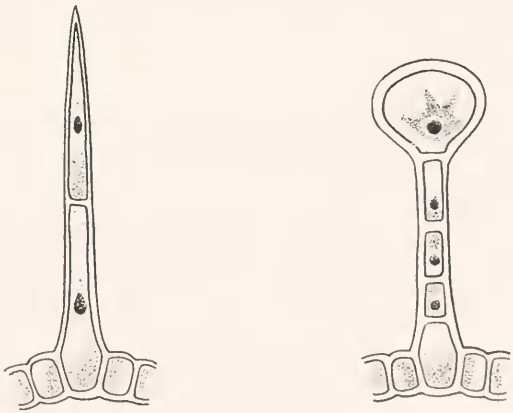

FIGURE 1. Diagrammatic longitudinal sections of leaf hairs of *Pelargonium*; left spine-shaped hair; right glandular hair.

left) were spinelike and the glandular hairs (Figure 1, right) appeared as spines with a rounded head.

Results.—In the present study G10 was found to be susceptible and G54 was found to be resistant to the twospotted spider mites with a significant difference at the 1% level using a leaf disc method (Table 1); this was based on egg production.

Table 1. A comparison of the mean numbers of eggs laid by twospotted spider mite on upper and lower leaf surfaces of cultivars 'G54' and 'G10'.

	Number of eggs laid	
	Upper	Lower
'G54'	1.62	1.33
'G10'	2.67	3.04

L.S.D. $_{.01} = 0.95$

Thickness of Cuticle and Epidermis: The upper cuticle of 'G54' was significantly thicker than the lower cuticle at the 1% level (Table 2). Similarly the upper cuticle of 'G10' was thicker than its lower cuticle. The upper cuticle of 'G54' was significantly thicker than the upper cuticle of 'G10' at the 1% level. The lower cuticles of these two cultivars are likewise significantly different in thickness. Similar differences were also observed in making comparisons of the mean thickness of epidermal cells in these two cultivars (Table 3).

Epidermal Hairs: No significant differences were observed in total number of hairs (Table 4) or spine-shaped hairs (Table 5) for any surface or cultivar comparisons.

Table 2. A comparison of the mean thickness of the cuticle on the upper and lower leaf surfaces of mite resistant ('G54') and mite susceptible ('G10') cultivars of *P.* x *hortorum*.

	Thickness of Cuticle (μ)		
	Upper	Lower	$\overline{X}$
'G54'	2.4	1.4	1.9
'G10'	1.5	1.0	1.2
$\overline{X}$	1.9	1.2	

Cultivars—°° L.S.D. $_{.01} = 0.15$
Surfaces—°° L.S.D. $_{.01} = 0.15$
Interaction—°° L.S.D. $_{.01} = 0.30$

TABLE 3. A comparison of the mean thickness of epidermal cells on the upper and lower leaf surfaces of mite resistant ('G54') and mite susceptible ('G10') cultivars of *P.* x *hortorum*

	Thickness of Epidermal Cell (μ)		
	Upper	Lower	$\overline{X}$
'G54'	18.0	11.2	14.6
'G10'	16.7	10.0	13.3
$\overline{X}$	17.3	10.6	

Cultivars—°° L.S.D. = 0.76
Surfaces—°° L.S.D. = 0.76
Interaction—N.S.

However, the number of glandular hairs on the lower surface was greater than the number of glandular hairs on the upper surface for both 'G54' and 'G10'. When 'G10' is compared to 'G54', the number of glandular hairs on both the upper and lower surfaces of the latter is significantly greater (Table 6) than the former.

DISCUSSION.—Morphological characteristics of two pelargonium cultivars, one resistant and one susceptible, to attack by the two-spotted spider mite were studied. The cuticles and epidermises of

TABLE 4. A comparison of the mean total number of hairs on 5.3 mm² of upper and lower leaf surfaces of mite resistant ('G54') and mite susceptible ('G10') cultivars of *P.* x *hortorum*. No significant differences recorded.

	Number of Hairs	
	Upper	Lower
'G54'	69.0	69.4
'G10'	65.4	70.6

TABLE 5. A comparison of the mean number of spine-shaped hairs on 5.3 mm² of upper and lower leaf surfaces of mite resistant ('G54') and mite susceptible ('G10') cultivars of *P.* x *hortorum*.

	Number of Hairs		
	Upper	Lower	$\overline{X}$
'G54'	64.7	58.5	61.6
'G10'	64.0	66.3	65.1

TABLE 6. A comparison of the mean number of glandular hairs on 5.3 mm² of upper and lower leaf surfaces of mite resistant ('G54') and mite susceptible ('G10') cultivars of *P.* x *hortorum*. No significant differences recorded.

	Number of Hairs		
	Upper	Lower	$\overline{X}$
'G54'	4.3	11.0	7.6
'G10'	1.4	4.3	2.8
$\overline{X}$	2.8	7.6	
Cultivars—**		L.S.D. $_{.01} = 1.57$	
Surfaces—**		L.S.D. $_{.01} = 1.57$	
Interaction—**		L.S.D. $_{.01} = 3.14$	

the resistant cultivar were significantly thicker than the cuticles and epidermises of the susceptible cultivar. Also cuticles and epidermises of the upper leaf surfaces were significantly thicker than their respective lower leaf surfaces in both cultivars.

The average thickness of G10 and G54 leaves used in this study was 237.5 μ while the average length of the chelicerae of adult female twospotted spider mites used in this study was 243 μ; the length of the chelicerae of the larval stage was 107.1 μ. It is not known to what depth the chelicerae of mites are able to penetrate leaf tissue; however, it does appear that larval mites do have difficulty penetrating the resistant leaf tissue. Perhaps resistance might be better studied with larval rather than with adult mites.

The total number of hairs and the number of spine-shaped hairs on the two cultivars and on the two leaf surfaces were not significantly different between the resistant and susceptible cultivars. However, the resistant cultivar had significantly more glandular hairs than did the susceptible cultivar. Also glandular hairs were significantly greater on the lower surface than on the upper surface of each cultivar. Both of these factors suggest that these glandular hairs may be related to resistance.

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2.0168 Leaf characteristics of spider mite resistant and susceptible cultivars of Pelargonium x hortorum.

Abstract.—Leaf characteristics of spider mite resistant and susceptible cultivars of *Pelargonium* × *hortorum*. A leaf disc technique was used to establish significant differences in mite fecundity between the cultivars of *Pelargonium* × *hortorum* Bailey; one was classified as resistant and the other susceptible to the twospotted spider mite, *Tetranychus urticae*. Thicker cuticular and epidermal layers and the presence of glandular hairs was associated with the resistant cultivar; it is suggested that larval survival may be a factor in resistance—*Kuo-koung Patricia Chang, Robert Snetsinger, and Richard Craig, College of Agriculture, The Pennsylvania State University, University Park, PA 16802.*

Descriptors: Acarina; *Tetranychus urticae*; *Pelargonium* × *hortorum*; plant resistance.